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Studies on two corn hybrids differing in their reaction to drought *

by

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Introduction : In order to determine the differences in certain physiological attributes associated with drought resistance, studies were made on two corn hybrids which were observed to differ in their reaction to drought under field conditions. Physiological characteristics studied included seed germination and rate of growth under different moisture stress, determination of transpiration loss, dry matter content and water requirement, osmotic concentration of cell sap and nitrogen fraction during hardening. These aspects of approach to the problem of drought hardiness in crop plants have been reviewed extensively by Ashton (1948) and Levitt (1956).

The present studies relate to two corn hybrids subjected to drought, artificially induced by moisture stress, in the laboratory and under green house conditions. The plants were grown at three levels of moisture in pots under controlled conditions and this enabled the study of physiological changes that take place during drought hardening. The early stages of seedling growth, which constitute one of the critical periods subject most to adverse environmental conditions, was alone taken into consideration. Haber (1938) reported that sweet corn strains of 15 days age were more heat tolerant than those of 29 days age. In another study on the heat tolerance of corn, young seedlings of 10 to 12 days old showed more heat resistance than 16 to 20 days old plants (Heyne and Laude 1940).

Materials and Methods : Two corn hybrids, viz., (Ky 27 × Ky 49)X (K155 × K10) B-S3-1-1-1-1-1 designated 'S' for drought susceptible, (K55×CI 64) X H30 designated 'R' for drought resistant were used in the study. These two hybrids were selected since their pedigrees suggested that a difference in drought response could be expected, with the former being more susceptible than the latter.

For seed germination under limited moisture conditions in the laboratory, D-mannitol solutions of 0, 8, 11 and 13 atmospheres of osmotic pressure were employed. The amount of D-mannitol and water to provide the different atmospheric tensions were calculated

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as per the formula given by Helmeric and Pfeifer (1954). Ten ml. of the appropriate solutions were added to the petri-dishes two to three times during the period of seven days of the experiment. Seeds were set on filter paper kept in petri-dishes of 10 cm. diameter after sterilisation and D-mannitol solution added. The seeds were covered with another filter paper and the dishes closed with lids and kept in the germinator at 25°C. On the seventh day the number of seeds germinated and length of coleoptile were recorded. Forty to 100 seeds in each variety were used for the test.

Further investigations were carried out by growing the plants in pots at 15%, 20% and 25% moisture levels. Corn seeds were planted at uniform depths of one inch below soil surface. Evaporation of moisture from soil surface was prevented by paraffin layer of half-inch thickness. Five seedlings were grown in each pot and there were four pots for each treatment, viz. 15%S, 15%R, 20%S, 20%R, 25%S and 25%R. Loss in transpiration was recorded every week and the loss made up by adding the appropriate quantity of water through glass tube resting on half inch layer of sand below the paraffin layer.

Rate of growth in all the plants was recorded at weekly intervals, commencing from 15th day after planting up to 10 weeks of growth. Sampling of plant material was done thrice at 5, 7 and 10 weeks growth for determining leaf water content, dry matter produced and soluble protein nitrogen content during hardening. Moisture and dry matter contents were determined with 10 gm. sample kept for drying at 75°C. Nitrogen determination was done by extracting the sap from leaf and stem in an osterizer and precipitating the protein with trichloroacetic acid. Kjeldhal digestion and distillation were carried out for determining the nitrogen content.

Osmotic concentration of cell sap was determined by cryoscopic method using Beckmann thermometer. Sampling of tops was done from 10 week old plants and sap extracted in a hydraulic press at 12,000 pounds per square inch of pressure for five minutes after pre-freezing the material in sealed plastic bags at — 12°C for 10 hours. The sap was centrifuged at 1600 revolutions per minute for five minutes. The freezing point of sap was determined and osmotic concentration calculated from the depression in freezing point.

Results and Discussion: *Studies with D-mannitol:* The germination of the hybrids was studied at 0, 8 and 11 atmospheres of

osmotic pressure in experiment 1 and at 0, 8 and 13 atmospheres in experiments 2 and 3. Chi-square tests were used to compare the germination percentages and the results are presented in table 1.

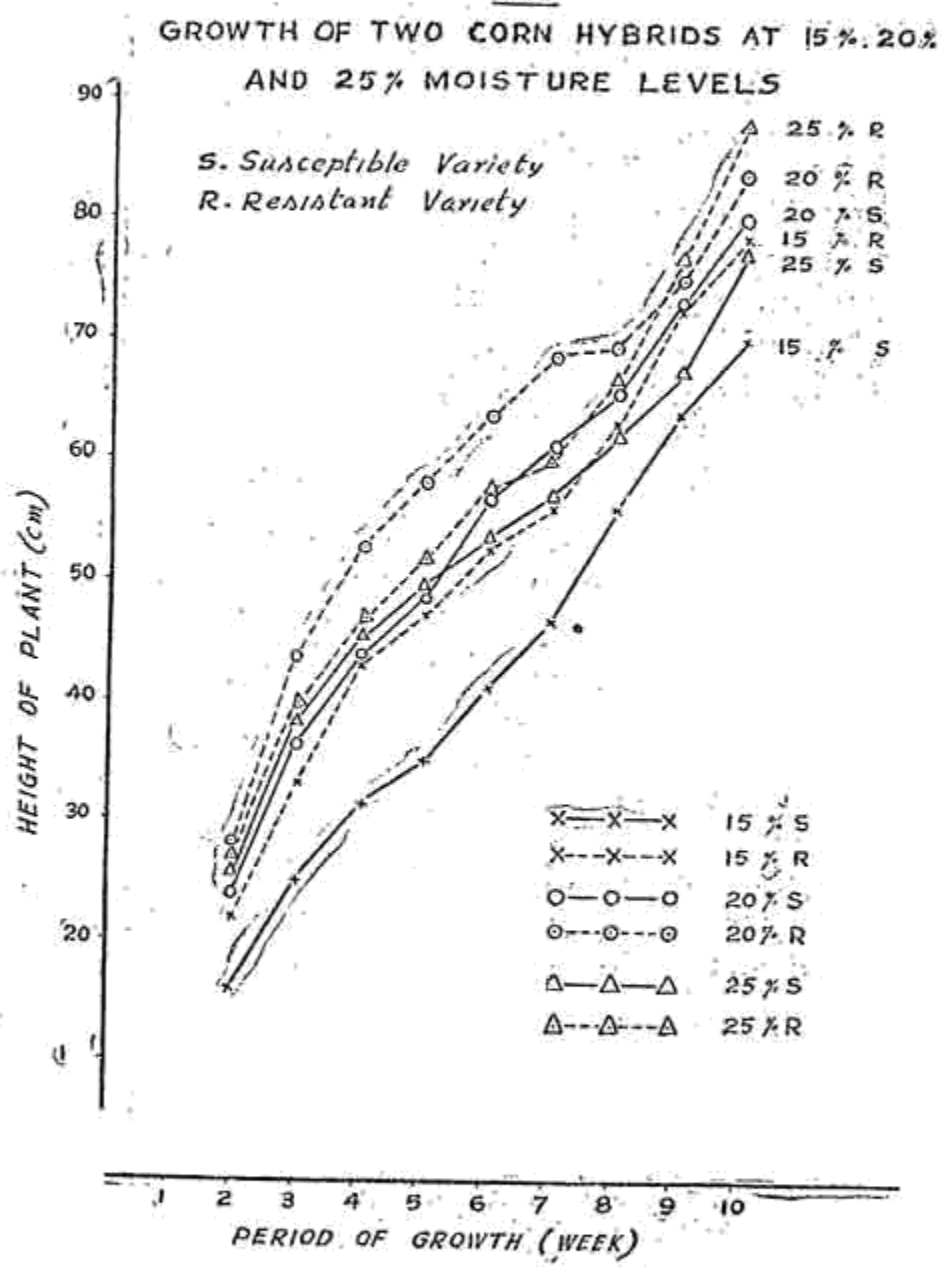
It is seen that significant differences are obtained at 11 atmospheres in experiment 1, 13 atmospheres in experiment 2 and at 8 and 13 atmospheres in experiment 3, the percentage germination in the resistant variety being significantly greater than that in the susceptible variety in these cases. But it is seen that the differences between coleoptile length in the resistant and susceptible types are not consistant. Hence it does not suggest that growth as recorded by the coleoptile length alone can be taken as an index of measurement to distinguish a susceptible variety from the resistant one. But the percentage of germination and survival of seedlings in D-mannitol may however, be considered a suitable index and a concentration of 13 atmospheres of osmotic pressure and a period of seven days appropriate for such a study in distinguishing a drought resistant type from the susceptible one.

According to Thimann (1954) D-mannitol is the best chemical found to limit water uptake in a plant without affecting the metabolic action of the plant. Studies with D–mannitol have been reported on winter wheat varieties by Helmeric and Pfeifer (1954) and Powel and Pfeifer (1956). They found that the use of D–mannitol solutions to test drought hardiness as a simple and easily repeated test which gives a relative measure of any difference among selections for their seedling character. The data from the preliminary studies on corn presented indicate that seed germination and relative rate of survival of the two hybrids are in agreement with their observed reaction to to soil drought under field conditions.

Rate of Growth : The height and rate of growth of the two corn hybrids in three moisture levels recorded at weekly intervals are given in Table 2 and Fig. 1. Germination and emergence were quick and pronounced in 25% moisture level followed by that in 20% moisture while germination in 15% moisture was slow. Emergence of seedlings in all pots was complete within seven days and the resistant hybrid showed superior emergence and taller growth than the susceptible one in the three levels of moisture (Fig. 2, pot Nos. 2, 4 & 6). The rate of growth (Fig. 1) indicates that there is marked difference between the two hybrids upto four weeks at 15% and 20% moisture levels. However, the differences in growth between the two hybrids over a period of 10 weeks do not approach the level of significance in the three levels of moisture though it is seen that at

25% moisture level, the difference is greatest and approaches near significance (Table 2). Hence it appears that early emergence and quick rate of growth up to a period of four weeks might be a criterion to distinguish the drought resistant type from a susceptible one and a moisture level of 25% or more might be suitable for the test.

FIG.1



Transpiration loss, dry matter content and water requirement : The total quantity of water transpired, the amount of dry matter produced and leaf water content at three stages of growth, viz. 5, 7 and 10 weeks are given in Table 3. In addition, the water requirement, that is the ratio between the amount of water transpired and the amount of dry matter produced is also furnished in the table.

The leaf water content does not show any marked difference between the two types but the indications are that the resistant type has less of leaf water content in the three samplings and at three levels of moisture. It is also evident from the data that the resistant variety has greater dry matter in the three stages of sampling. In the ultimate analysis of this aspect of studies it is the physiologic water requirement that deserves due consideration. It is seen that the resistant type has lower water requirement than the susceptible one. The differences are wider at greater moisture stress and in the early stages of growth, at 5 and 7 weeks of growth. Though the studies have been limited to 10 week old seedlings, an attempt has been made to correlate the water requirement of the two corn hybrids with their relative reaction to drought. The indications are that the resistant hybrid has more of dry matter content and less of water requirement.

Soluble protein nitrogen content : The data on soluble protein nitrogen content of stem and leaf samples in the two corn hybrids determined at 5, 7 and 10 weeks of growth (Table 4) show that there is a gradual increase in the nitrogen content as the moisture stress increases in all the three samples and the resistant variety shows increased nitrogen content. The differences are, however, narrow at higher moisture levels.

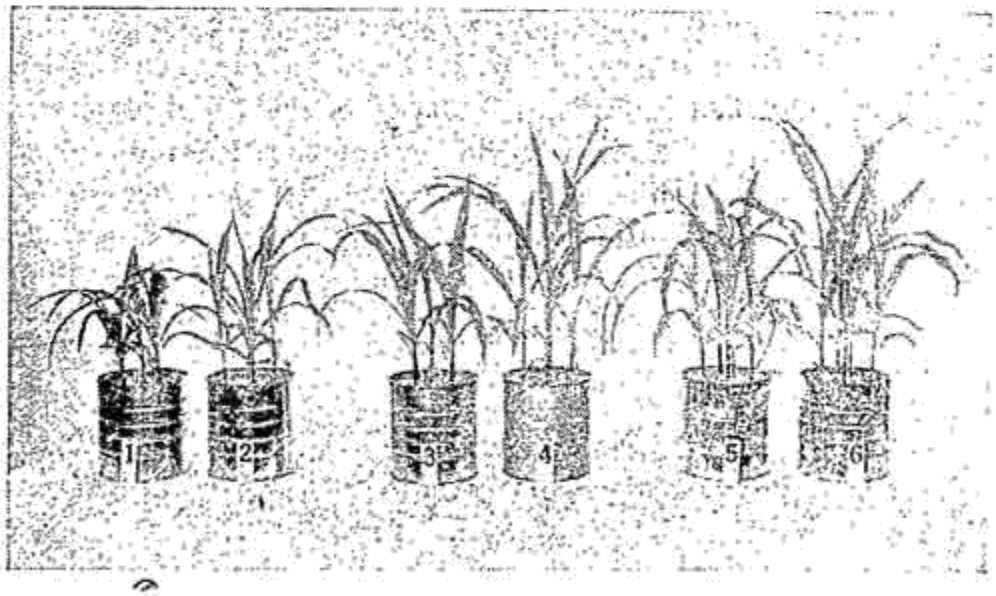


FIG. 2

Drought resistant (R) and susceptible (S) corn varties grown at 15%, 20% and 25% moisture levels showing difference in growth (5 weeks after planting).

1. 15% S; 2. 15% R; 3. 20% S; 4. 20% R; 5. 25% S; 6. 25% R.

It was suggested by Nightingale and Farnham (1936) that water deficit interferes with protein synthesis in plants. Clements (1937) (quoted by Kramer 1949) found a high level of nitrogen metabolism in plants that were grown with deficient soil moisture. Black (1957) has also pointed out that the nitrate content of plants is high under conditions of drought. The data obtained from this preliminary study indicate that there is an increase in nitrogen content as the soil moisture stress becomes greater and the resistant variety has slightly increased nitrogen content

Osmotic pressure of cell sap : The freezing point depression and osmotic pressure of cell sap from tops of the two corn hybrids grown at 15%, 20% and 25% moisture levels and sampled when ten weeks old are given in Table 5. The results show that there is very little difference between the resistant and susceptible hybrid in their osmotic concentration. But the osmotic pressure of the sap from plants grown in 15% and 20% moisture levels is greater than that of the plants grown at 25% moisture level.

Levitt (1956) has tabulated the increased osmostic value in species on exposure to drought and has stressed that osmotic value is definitely a factor in drought hardiness, though not the deciding one. Variation in osmotic pressure is attributed partly to changes in carbohydrate content and partly to changes in moisture content. In the present studies no varietal difference has been observed but there is an increase in value of more than two atmospheres in plants exposed to moisture stress. It is quite likely that the difference in osmotic value is due to variation in leaf water content as seen in Table 3. The data show that the plants grown at 15% moisture level have 4% less of water content as compared to that from 25% moisture level.

Summary : Two corn hybrids differing in their reaction to drought in field were tested in the early stages of seedling growth under laboratory and green house conditions for certain physiological attributes associated with drought hardiness.

D-mannitol solutions of 0, 8, 11 and 13 atmospheres of osmotic pressure were used to compare the germination and relative rate of growth upto a period of seven days. Significant differences in germination and survival were obtained at 13 atmospheres in majority of the cases.

Growth rates of plants at 15%, 20% and 25% moisture levels for a period of 10 weeks indicated that at 25% moisture level the difference in growth was greatest and approached near significance. At 15% and 20% moisture levels, the resistant variety showed superior emergence and quick rate of growth upto four weeks.

The resistant hybrid showed less of leaf water content and hardening was found to decrease the leaf water content. In addition, the resistant hybrid had more of dry matter content and less of water requirement.

There was a gradual increase in soluble protein nitrogen in the shoot as the moisture stress increased and the resistant hybrid showed increased values. The differences became narrow at higher moisture levels.

No varietal difference in osmotic pressure of cell sap from tops was obtained but there was an increase of more than two atmospheres on exposure to increased moisture stress. This might be due to decreased leaf water content under drought hardening.

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TABLE 1.

Average coleoptile length and percentage germination of two corn hybrids at thres levels of atmospheric tensions compared with distilled water (control).

Experiment	Variety	Atmospheric Tension of Mannitol Solutions							
		0		8		11		13	
		Colepotile length mm.	% Germination	Coleoptile length mm.	% Germination	Coleoptile length mm.	% Germination	Coleoptile length mm.	% Germination
1	S	5·12	96	1·37	80	0·58	58	...	...
	R	5·91	100	1·90	90	0·83	86	...	...
2	S	5·19	90	2·72	75	...	...	0·80	45
	R	4·91	98	1·94	80	...	...	0·83	70
3	S	3·79	92	2·05	72	....	...	0·65	44
	R	3·12	95	1·25	87	...	...	0·66	71

Chi-square values for comparing the germination of resistant and susceptible varieties

SITUATION	X^2, 1 D/F	SIGNIFICANCE
Experiment 1 — 0 Atmospheres	1·02	$P > 0·25$
,, 8 ,,	1·96	$P > 0·10$
,, 11 ,,	9·72	$P < 0·005$*
Experiment 2 — 0 ,,	1·92	$P > 0·10$
,, 8 ,,	0·09	$P > 0·75$
,, 13 ,,	5·11	$P < 0·025$*
Experiment 3 — 0 ,,	0·74	$P > 0·25$
,, 8 ,,	6·91	$P < 0·010$*
,, 13 ,,	14·03	$P < 0·005$*

* Significant

Height of two corn varieties grown in pots at 15%, 20% and 25% moisture levels. (Average of four plants).

Treatments	Height of plant at weekly intervals (cm.)									$b_s - b_r$	t
	2 weeks	3 weeks	4 weeks	5 weeks	6 weeks	7 weeks	8 weeks	9 weeks	10 weeks		
15% S ...	16·22	25·06	31·54	35·09	41·41	46·81	56·08	63·98	70·28		
										— 0·003	...
15% R ...	21·95	32·78	43·06	47·01	52·63	56·08	63·43	72·43	78·53		
20%1S ...	23·82	36·28	43·63	48·44	56·70	61·13	65·40	72·78	80·13		
										+ 0·479	0·74
20% R ...	28·21	43·83	52·52	58·09	63·79	68·68	69·25	75·15	84·15		
25% S ...	25·62	38.19	45·29	49·22	53·80	56·83	62·00	67·43	77·65		
										— 1·122	1·78
25% R ...	27·41	39·75	46·54	51·86	56·70	60·26	66·63	76·68	87·98		
										(P^2 = ·09)	

TABLE 3.

Quantity of water transpired, amount of dry matter produced, leaf water content and water requirement in two corn varieties grown at 15%, 20% and 25% moisture. (Average of four plants).

Treatments	Quantity of water transpired (gm)			Amount of dry matter produced (gm)			Leaf water content (%)			Water requirement: Amount of water transpired / Amount of dry matter produced		
	5 weeks	7 weeks	10 weeks	5 weeks	7 weeks	10 weeks *	5 weeks	7 weeks	10 weeks	5 weeks	7 weeks	10 weeks *
15% S ...	61·12	149·02	415·92	0·2378	0·5289	3·3720	83·42	85·86	81·87	257·02	281·75	123·35
15% R ...	86·60	194·50	505·80	0·4471	0·9309	4·6353	83·22	85·20	78·79	193·39	208·94	109·92
20% S ...	118·78	252·68	674·80	0·4588	0·9400	4·9739	86·94	86·00	82·02	258·89	268·81	135·67
20% R ...	131·86	278·26	686·76	0·6070	1·4925	5·7110	86·66	83·87	80·23	217·23	186·44	120·25
25% S ...	126·40	246·40	701·70	0·5460	0·7452	5·3960	87·99	86·21	84·54	231·50	330·65	130·04
25% R ...	120·62	250·52	728·62	0·5438	0·8815	6·7840	86·78	85·29	82·82	221·81	284·19	107·40

* Includos weight of root.

TABLE 4.

Soluble protein nitrogen content in tops of two corn varieties grown at 15%, 20% and 25% moisture levels.

Treatments	Nitrogen content at three stages of crop growth %		
	5 weeks	7 weeks	10 weeks
15% S	0·0574	0·0139	0·0748
15% R	0·0809	0·0226	0·1035
20% S	0·0305	0·0113	0·0505
20% R	0·0435	0·0139	0·0696
25% S	0·0200	0·0096	0·0522
25% R	0·0226	0·0131	0·0722

TABLE 5.

Osmotic pressure of cell sap from tops of two corn varieties grown at 15%, 20% and 25% moisture levels. (Cryoscopic determination)

Treatments	Freezing point depression °C	Osmotic pressure (atmospheres)
15% S	1·0497	12·638
15% R	1·0468	12·605
20% S	1·0320	12·425
20% R	1·0568	12·724
25% S	0·8859	10·666
25% R	0·8477	10·206

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